Express Mail Label No.: EV679164101US

Attorney Docket No.: 3087.00024

IN THE CLAIMS:

1. (Currently amended) An antibody microarray screen comprising: a substrate; monoclonal and polyclonal antibodies that are purified immunoglobulins immunoglobins, wherein said antibodies are spotted on predetermined positions on said substrate; and fluids unprocessed for immunoglobulin isolation, wherein said unprocessed fluids with higher amounts of proteins than said purified immunoglobulin proteins to compensate non-immunoglobulin proteins in said unprocessed fluids are spotted on said predetermined positions on said substrate.

- 2. (Original) The antibody microarray screen according to claim 1, wherein said antibodies detect proteins selected from the group consisting of drug-metabolizing enzymes and proteins functionally related with said drug-metabolizing enzymes.
- 3. (Original) The antibody microarray screen according to claim 2, wherein said drug metabolizing enzyme is cytochromes P450.
- 4. (Original) The antibody microarray screen according to claim 2, wherein said proteins functionally related with said drug-metabolizing enzyme are selected from the group consisting of mitochondrial proteins, apoptosis-related proteins, anti-oxidant proteins, oxidative stress proteins, and intracellular protein degradation proteins.
- 5. (Original) The antibody microarray screen according to claim 1, wherein said substrate includes a hydrogel (polyarylamide-based) coating.
- 6. (Currently amended) The antibody microarray screen according to claim 1 and determining optimal spotting concentrations of IgG by further

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comprising steps of (a) hybridizing the slides with labeled secondary immunoglobulins, (b) scanning and quantitating signal strength of each spot, and (c) selecting optimal concentrations of IgG using %visible IgG spots.

- 7. (Original) The antibody microarray screen according to claim 1, wherein said fluids are selected from the group consisting of ascites fluids, hybridoma culture medium, and anti-sera.
- 8. (Currently amended) An antibody microarray screen comprising: a substrate; polyclonal antibodies as purified <u>immunoglobulins</u> <u>immunoglobulins</u>, wherein said antibodies are spotted on predetermined positions on said substrate; and anti-sera <u>with higher amounts of proteins than said purified immunoglobulin proteins to compensate non-immunoglobulin proteins in said <u>unprocessed fluids are</u> spotted on said predetermined positions on said substrate.</u>
- 9. (Original) The antibody microarray screen according to claim 8, wherein said polyclonal antibodies detect proteins selected from the group consisting of drug-metabolizing enzymes, cytochromes P450, and oxidative stress proteins.
- 10. (Original) The antibody microarray screen according to claim 8, wherein said substrate includes a hydrogel (polyarylamide) coating.
- 11. (Currently amended) The antibody microarray screen according to claim 8 and determining optimal spotting concentrations of IgG by further including comprising steps of (a) hybridizing the slides with labeled secondary immunoglobulins, (b) scanning and quantitating signal strength of each spot, and (c) selecting optimal concentrations of IgG using %visible IgG spots.

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immunoglobins.

12. (Currently amended) An antibody microarray screen comprising: a

substrate; monoclonal antibodies as purified immunoglobulins, immunoglobin,

wherein said antibodies are spotted on predetermined positions on said

substrate; ascites fluid spotted on said substrate; and hybridoma culture media

spotted on said substrate, wherein said ascites fluid and hybridoma culture

media with higher amounts of proteins than said purified immunoglobulin

proteins to compensate non-immunoglobulin proteins in said unprocessed fluids

are spotted on predetermined positions on said substrate.

13. (Original) The antibody microarray screen according to claim 12,

wherein said monoclonal antibodies detect proteins selected from the group

consisting of drug-metabolizing enzymes, cytochromes P450, and oxidative

stress proteins.

14. (Original) The antibody microarray screen according to claim 12.

wherein said substrate includes a hydrogel (polyarylamide) coating.

15. (Currently amended) The antibody microarray screen according to

claim 12 and determining optimal spotting concentrations of IgG by further

comprising steps of (a) hybridizing the slides with including labeled secondary

immunoglobulins, (b) scanning and quantitating signal strength of each spot, and

(c) selecting optimal concentrations of IgG using %visible IgG spots.

immunoglobins.

16. (Original) A method of manufacturing an antibody microarray

comprising the step of spotting more than a single concentration of antibodies on

a microarray substrate to increase the number of up-regulated protein detection.

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17. (Original) The method according to claim 16, wherein the antibody concentration is more than 5 µg/ml lgG.

18. (Original) An internal control molecule for use in an antibody

microarray comprising a protein, wherein said protein is unexpressed in the array

sample for normalization of focused (non-global) array data.

19. (Original) The internal control molecule according to claim 18,

wherein said protein is selected from the group consisting of a Flag protein and a

non-mammalian protein.

20. (Original) The internal control molecule according to claim 18,

wherein the internal control molecule is used to compare the expression ratio of

house-keeping proteins to select housekeeping genes by determining any

difference between the control and experimental samples.

21. (Original) A method of determining optimal spotting concentrations of

IgG comprising the steps of: (a) spotting increasing concentrations of IgG on

microarray slides; (b) hybridizing the slides with secondary IgG with a detectable

signal; and (c) scanning and quantitating signal strength of each spot and

selecting optimal concentrations of IgG.

22. (Original) A method to increase a detectable signal with microarray

analysis comprising the steps of using an intensive molecular signal, wherein the

intensive molecular signal is produced by conjugation of a dye and a reporter

molecule to a protein whereby interference of IgG binding to a protein is created.

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23. (Original) The method according to claim 22, wherein the intensive molecular signal is produced by conjugation of a dye and a reporter molecule to a protein to the extent that interference of Coomassie blue stain binding to the protein is created.

- 24. (Original) The method according to claim 22, wherein the intensive molecular signal is used for antibody microarrays.
- 25. (Original) The method according to claim 22, wherein the intensive molecular signal is used for protein microarrays.
- 26. (Original) A method to increase a detectable signal with microarray analysis comprising the steps of: conjugating of a dye and a reporter molecule to a protein; and creating interference of an IgG molecule binding to the protein.
- 27. (Original) A method of producing antibody microarrays comprising the steps of spotting antibodies for Phase I and II drug metabolizing enzymes and proteins functionally related with the drug-metabolizing enzymes on a microarray substrate.
- 28. (Original) The method according to claim 27, wherein the proteins functionally related with drug-metabolizing enzyme are selected from the group consisting of mitochondrial proteins, apoptosis-related proteins, anti-oxidant proteins, oxidative stress proteins, and intracellular protein degradation proteins.
- 29. (Original) The method according to claim 27, wherein the targeted drug-metabolizing enzyme antibody microarray includes an internal control to be used for data normalization.

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30. (Original) The method according to claim 29, wherein the internal control is a Flag protein.